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Determination of Minimum Number of Frustule for Identification of Diatoms in Telaga Cebong, Dieng, Central Java, Indonesia

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Abstract

Diatoms belong to the division of Bacillariophyta; their cell walls composition of silica (SiO_2) was the primary characteristic of their structure. Diatoms are good for water quality assessment and paleolimnological analysis. The determination number of frustule which has to be counted for diatom is one of the critical steps in process identification. Several studies usually use a range starting from 100-600 frustules, which refers to the "fixed count" method, that is time-consuming. Research about the minimum frustules number that is acceptable for water character in Indonesia has been limited studied. This research aim is to determine the minimum number of frustules need to identify the diatom assemblage, especially at Telaga Cebong, Dieng, Central Java, Indonesia. The sediment samples were taken vertically with D'Section corer until a depth of 200 cm sediment samples then sliced every 10 cm. The labwork activites consisted of digestion, preparation, enumeration, and identification of diatom species with the numberof frustules of 100,200, 300, 400, 500, and 600. The diatom data are analyzed with the formula for the maximum efficiency value. Based on 19 sediment samples from different sediment layers, the maximum efficiency value obtained was above the range of 0.85 to 0.92. The number of identified species remained stable after a minimum of 300 frustules. Therefore, at least 300 frustules are needed for identification of diatom for paleoreconstructions at Cebong Lake.

Keywords: Diatom, Minimum frustule, Paleolimnology, Telaga Cebong Dieng.

1. Introduction

Diatoms play an essential role in the aquatic food webs as a producer. Diatom cell walls composed from silica, therefore, it can be fossilized in sediments and used for paleolimnological analysis (Soeprobowati *et al.*, 2016). Diatoms with the highest abundance in waters have a sensitive response to changes in the ecosystem, such as light, temperature, salinity, and pH (Leterme *et al.*, 2013). In the analysis of diatoms as an indicator of water quality or as a paleo reconstruction, the identification of diatoms is an important step for the interpretation result. The diatom identification process previously required several stages called a protocol consisting of digestion, preparation, and identification (Soeprobowati *et al.*, 2016). The number of frustules must be determined as this is the main point of all standards and affects the assessment status of an environment by associating the diatom taxa that are abundantly represented (Brabcova´ *et al.*, 2017).

The general calculation method that is often used on regular protocol made by Battarbee (1986) a fixed number of 300 to 600 frustules is designed to characterize the relative abundance which are often used to assess river water quality. The fixed number make the calculations efficient and affordable (Kelly *et al.*, 2008). Round (1993) used 100 diatom frustules in identification for epiphytic diatoms. Meanwhile, Bates

& Newall (2002) suggest with 200 diatom frustules can be the ideal number to characterize the diatom assemblage for water quality asssessment. Ruhland (2003) identified at least 350 frustules in paleo reconstruction studies. However, this study focuses on locations that have moderate temperatures. Tropical water conditions affects the diversity of diatoms. Research conducted by Soeprobowati *et al.* (2016) determined that at least 300 diatom frustules had been able to be used to analyze the paleolimnology for Lake Rawapening, Indonesia

This study aims to determine the minimum amount of frustules in diatom analysis, especially at the diatom identification stage for paleo reconstruction purposes at Telaga Cebong, Dieng, Wonosobo, Central Java, Indonesia.

2. Methods

Telaga Cebong is one of the volcanic lakes located in the Dieng Plateau area about 2,000 meters above sea level, in Sembungan Village, Kejajar District, Wonosobo Regency, Central Java, Indonesia. The position of this lake is between Mount Pakuwojo (3,395 m asl), Seroja (1,300 m asl) and Sikunir (2,306 m asl) (Setiawan, 2012). This lake originated from an ancient crater. Telaga Cebong was selected for the research site because this lake is classified in the category of eutrophic lakes due to changes in land use from forest to potato farming land (Sudarmadji, 2015).

Two hundred centimeters sediment samples were taken using the D'Section corer. The sediment core was then sliced at every 10 centimeters. First stage is extraction aim to separate the organic materials. In this research the sediment sample is heating with 50 ml of 10% HCl to remove $CaCO_3$ contained in the sediment (Peabody, 1977). The sample is heated using 50 ml of 10% H_2O_2 the function is to separate the organic material contained in the sediment (Soeprobowati *et al.*, 2016). The second stage is mounting, the sample is dripping on a coverslip which is then closed and glued using Naphrax. The last stage is the identification and enumeration of diatoms. Observation using a microscope with 1,000x magnification. A total of 600 valves were counted per sample with a count of every 100 frustules (i.e. 100, 200, 300, 400, 500, 600). The step of determining the minimum efficiency value of diatom frustule is carried out using an accumulation curve applied to identify the minimum number of frustules to be calculated to achieve maximum efficiency. The maximum efficiency reflects the probability of a new diatom species to be found in each identification step (Pappas & Stoermer 1996; Bates & Newall 2002). The formula (Bates & Newall, 2002; Soeprobowati *et al.*, 2016):

$$\text{Maximum efficiency} = \frac{\text{Number of Species}}{\text{Number of Individual}}$$

3. Results and Discussion

Based on the results of the calculation of the minimum value of efficiency based on the diatom population, it can be seen in Figure 1 the form of a graph on the X-axis, is the number of frustules calculated and the Y-axis is the population. The lowest value on the graph of 0.8 was found in the 150 cm layer with a count of 100 individuals. While the average value of the largest efficiency is obtained at a layer of 70 cm with a minimum value of 0.95 efficiencies and a range of 0.89-0.95. Based on the graphic data, it can be seen that the efficiency value increases with the increasing in the number of individuals. This value is obtained based on the calculation of the logarithmic formula of the ratio between the number of species and the number of individuals, so a fixed number is not needed. The number of minimum values, species versus the number of individuals is logarithmic will be the starting point. Instead of using a predefined fixed amount.

In general, the minimum value of efficiency has an increasing trend start from the number of individuals counting from 100 to 400 with a significant amount. Next calculation, at the counting of 500 and 600 individual numbers, several samples were observed on the graph that continued to experience an upward trend or remained linear. Based on research conducted by Soeprobowati *et al.*, (2016) to obtain the minimum number of frustule, the number of calculated values with the highest and most stable efficiency values in all observed layers.

Based on the average trend in the graph, it can be seen that for the diatom identification process carried out at Cebong Lake, Dieng, Central Java, Indonesia at least 300 individual diatoms are needed with an average efficiency value of 0.92 (Figure 1). The selection of 300 frustules as the number specified in the diatom identification activity was chosen because the efficiency value of each layer has reached a value of more than 0.85, as stated by Soeprobowati *et al.*, (2016).

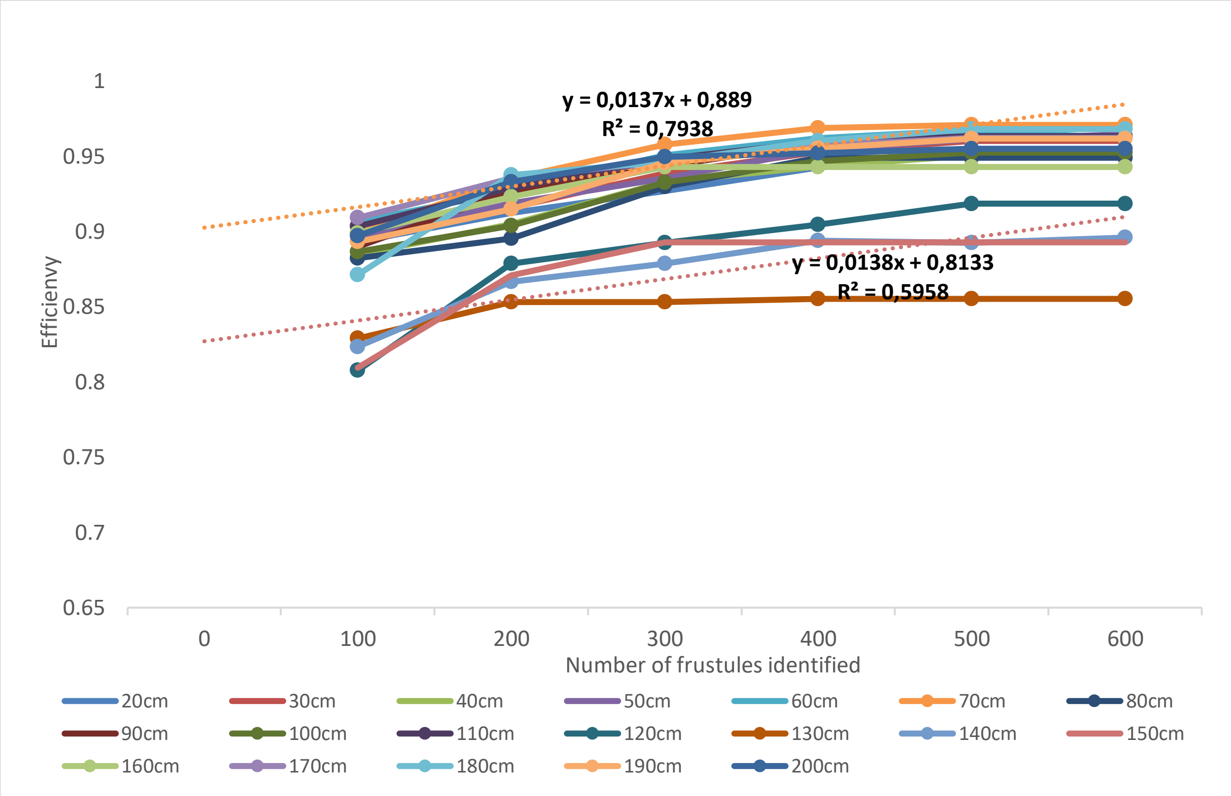

Figure 1. The efficiency value from the number of frustules identified from the 200 cm sediment samples depth in Telaga Cebong Dieng.

The determination of the minimum number of 300 diatom frustules is close to the statement of Battarbee (1986) which provides a determination of the recommended amount in the process of calculating diatom frustule, which is 300 to 600, however, this study is suitable for use in temperate climate. Indonesia with tropical climate conditions has different characteristics than the temperate regions. The Dieng Plateau has a characteristic climate and conditions that are different from other plains in Indonesia. Setyowati (2009) stated that the Dieng area includes Cebong Lake has a fairly high geographical location above 2,000 meters above sea level and cold or low temperatures as well as volcanic morphology and various symptoms such as active craters, fertile soil, crater lakes, or puddles, resulting in distinctive climatic and ecological conditions. This characteristic can be a reason why the number of frustule determinations in Cebong Lake has similarities with waters from temperate climate.

It can be seen from Figure 2 that the number of 100 to 300 frustules was tend to increase in the number of species was very significant in each layer sample, in contrast to the discovery of new species in the subsequent calculation of the number of frustules, starting from 300, 400, 500, and so on. In the number of 400 -600 frustules, many species found did not experience significant changes, even in some layers, no new species were found, which means that the discovery of new species in the count of 400 individuals or more is only incidental. This is also found in Lake Rawapening Cental Java (Soeprobowati *et al.*, 2016) which states that in the 300 frustules counted, the number of diatom species found to be stable. This is similar to at the Telaga Cebong that the number of 300 frustules the number of diatoms found did not experience much increase and tended to be stable. So that this amount can be used as a minimum value of efficiency that is suitable for use in the waters of Telaga Cebong, Dieng, Central Java, Indonesia. Meanwhile, according to Bates & Newall (2002) at least to carry out biomonitoring activities a calculation level is assumed to reach 80% efficiency to be accepted. The level of efficiency required depends on the use of data.

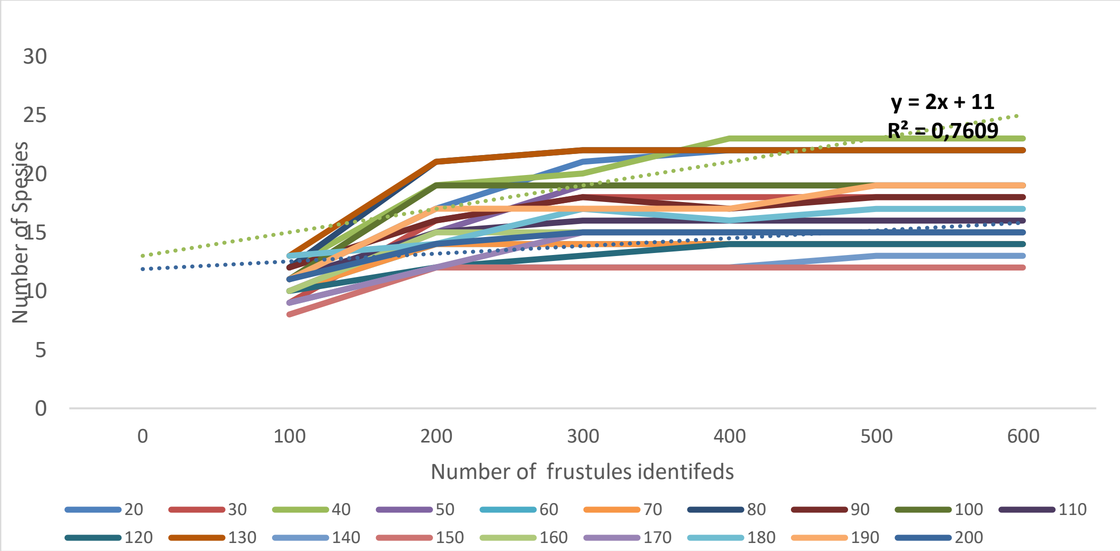

Figure 2. Number of species of diatoms from 200 cm sediment samples depth from Telaga Cebong Dieng

Each lake has its own water characteristics. Even in areas with the same weather conditions and characteristics, waters will still be able to have different dominant species. This is based on research conducted by Soeprobowati et al., (2016) at Rawapening Lake, the minimum number of frustule values needed for analysis activities in the lake is 300 frustules for paleolimnological analysis activities.

The dominant species found from Telaga Cebong Dieng from the genus of *Achnantidium* (Figure 3), *Denticula* (Figure 4), *Encyonema* (Figure 5), *Eunotia* (Figure 6), *Fragillaria* (Figure 7), *Gomphonema* (Figure 8), *Navicula* (Figure 9), *Nitzschia* (Figure 10), and *Staurosira* (Figure 11). Some species from those genera are potential for water quality indicator, such as *Fragillaria* and *Staurosira* usually find in the neutral to alkali pH (Pestryakova *et al*, 2018). *Achnanthidium minutissimum* is bioindicator for acid water (Stevenson & Bahls, 1999; Soeprobowati *et al.*, 2018), but Falasco *et al.* (2021) stated that this species also able to found in the oligotrophic to eutrophic.

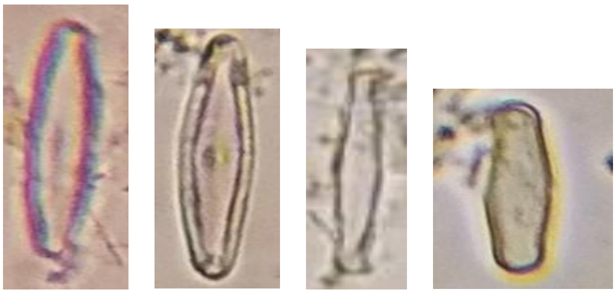

Figure 3. *Achnanthidium* sp from at Telaga Cebong, Dieng, Central Java.

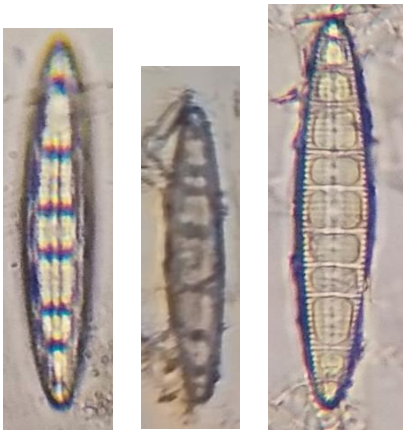

Figure 4. *Denticula* sp from Telaga Cebong from at Telaga Cebong, Dieng, Central Java

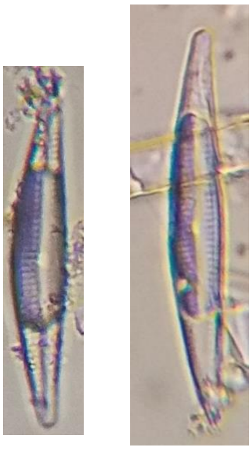

Figure 5. *Encyonema* sp from Telaga Cebong from at Telaga Cebong, Dieng, Central Java

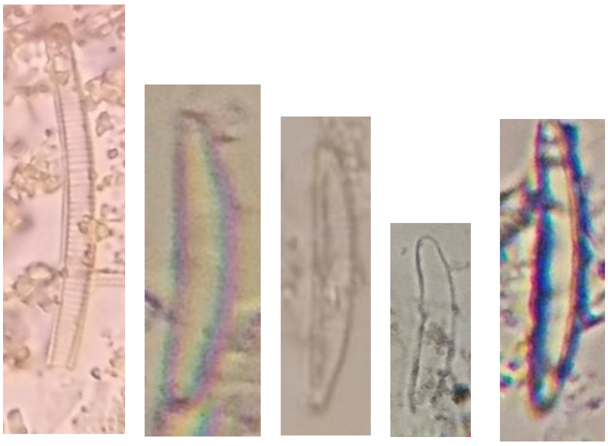

Figure 6. *Eunotia* sp from Telaga Cebong from at Telaga Cebong, Dieng, Central Java

Figure 7. *Fragillaria* sp from Telaga Cebong from at Telaga Cebong, Dieng, Central Java

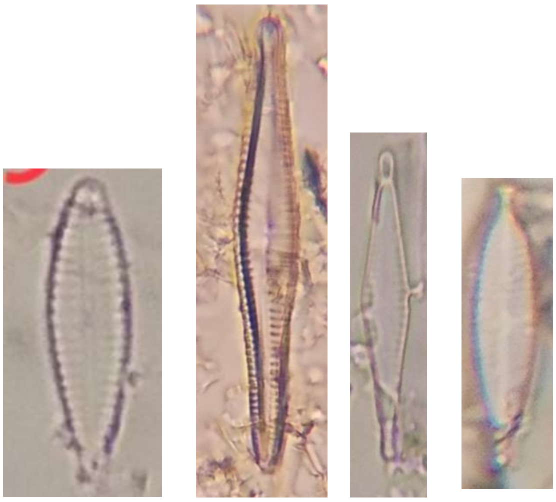

Figure 8. *Gomphonema* sp from Telaga Cebong from at Telaga Cebong, Dieng, Central Java

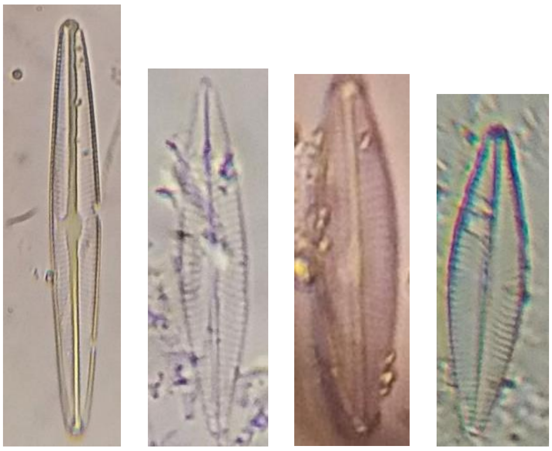

Figure 9. *Navicula* sp from Telaga Cebong from at Telaga Cebong, Dieng, Central Java

Figure 10. *Nitzschia* sp from Telaga Cebong from at Telaga Cebong, Dieng, Central Java

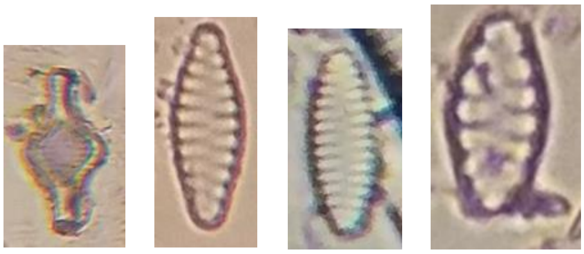

Figure 11. *Staurosira* sp from Telaga Cebong from at Telaga Cebong, Dieng, Central Java

4. Conclusion

The maximum efficiency of diatoms goes up 0.85 at a minimum of 300 frustules. The minimum number of diatom frustules recommended for use in diatom identification activities at Telaga Cebong, Dieng, Central Java, Indonesia is 300 individuals. Number of frustules in this research can be the reference for the counting number of diatom in the lake especially for the tropical climate region around Indonesia.

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